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Clinical and Cytogenetic Remission Induced by Interferon-α in a Patient With Chronic Eosinophilic Leukemia Associated With a Unique t(3;9;5) Translocation

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A patient with chronic eosinophilic leukemia and a unique complex chromosomal translocation 46,XY,t(3;9;5)(q25;q34;q33) who had elevated blood interleukin-5 is reported. Complete cytogenetic remission was induced by interferon-α after treatment failure with corticosteroids and cytotoxic drugs. Severe cardiopulmonary symptoms due to hypereosinophilia and thromboembolic complication improved dramatically in the first 6 months of interferon therapy. Since it is known that the gene encoding for interleukin-5 resides on the long arm of chromosome 5, it may be possible that the chromosomal translocation in our patient resulted in overproduction of this cytokine, and our findings may be helpful for understanding the pathogenesis of this disorder. Am. J. Hematol. 58:137–141, 1998.

Key words: chronic eosinophilic leukemia; interleukin-5; interferon-alpha; complex translocation; t(3;9;5); cytogenetic remission

INTRODUCTION

Eosinophilia can occur in several types of malignant hemopoietic disorders, such as eosinophilic leukemia, subtypes of acute myeloblastic leukemia and lymphoblastic leukemia, myeloproliferative disorders, and myelodysplastic syndromes. Among these, chronic eosinophilic leukemia is a rare hematological malignancy that is difficult to distinguish from idiopathic hypereosinophilic syndrome (HES). The diagnosis is made by demonstrating cytogenetic abnormalities in bone marrow progenitor cells. Translocations with 5q breakpoints between 5q31 and 5q33 have been associated with chronic eosinophilic leukemia [1] and it could be postulated that the location of genes for various cytokines, including interleukin-3 (IL-3), interleukin-5 (IL-5), and granulocyte/macrophage colony-stimulating factor (GM-CSF), in this region of chromosome 5 is relevant in the pathogenesis. Here we report on a case of chronic eosinophilic leukemia with a unique previously undescribed chromosomal abnormality and an increased blood level of IL-5. Treatment with interferon-α (IFN-α) resulted in long-term survival and complete cytogenetic remission, as has been seen with chronic myeloid leukemia (CML) [2].

CASE REPORT

A 26-year-old man was referred to our hospital in May 1990 with an intractable cough unresponsive to glucocorticoids that had been administered under a diagnosis of Loeffler's syndrome at a local hospital. Physical examination revealed a grade III/VI apical systolic murmur. There was no hepatosplenomegaly. Laboratory studies

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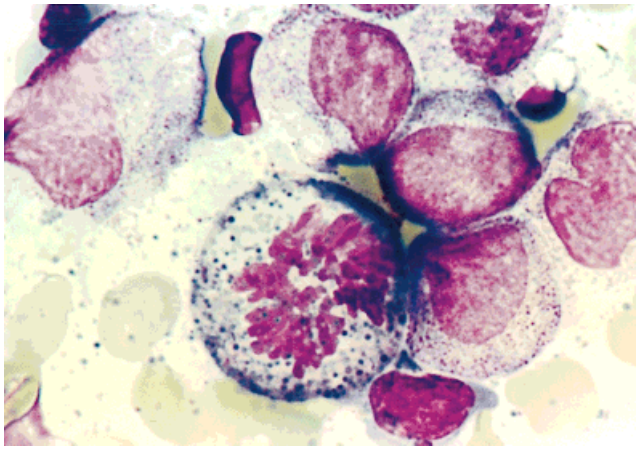

Fig. 1. Appearance of eosinophils in the bone marrow: the presence of eosinophilic granules in the background of the metaphase directly supports clonal eosinophilic proliferation (May-Grunwald-Giemsa stain, ×1,000).

revealed a white blood cell count of 13×10^9 1 with 31% eosinophils. The hemoglobin was 14.2 g/dl and the platelet count was 187×10^9/1. Bone marrow aspiration was hypercellular with a marked predominance of eosinophils and the cells undergoing mitosis frequently had eosinophilic granules, but there was no excess of blasts (Fig. 1). Leukocyte alkaline phosphatase was 280 (normal range, 170 to 375). Cytogenetic studies performed on bone marrow cells after culture for 24 h without the addition of mitogens revealed a 46,XY,t(3;9;5)(q25;q34;q33) in all of the 30 mitoses analysed. High molecular weight DNA extracted from the patient's bone marrow cells was employed for Southern blot analysis and excluded rearrangements of bcr-abl, T-cell antigen receptor, and platelet-derived growth factor receptorβ (PDGFRβ) gene (Fig. 2). Serum vitamin B12 was 14,300 ng/l (normal: 300 to 1,000 ng/l), and the serum IgE level was normal. Serum IL-3 and GM-CSF levels were not elevated before treatment. IL-5 was measured using a sandwich type enzyme immunoassay (Immunotech, Marseille, France) according to the kit procedure and increased to 39.2 pg/ml in the blood sample obtained after the initiation of IFN-α in April 1994. Using this kit, the upper limit of IL-5 serum level in healthy subjects is less than 7 pg/ml. Stool samples were negative for ova and parasites. Cardiac catheterization performed prior to treatment with IFN-α revealed normal coronary arteries with a dip and plateau pattern of the left ventricular pressure curve, severe mitral regurgitation, and moderate tricuspid regurgitation. The pulmonary capillary wedge pressure was 32 mm Hg.

The patient's course of treatment and changes in laboratory parameters are presented in Figure 3. Initial treatment with prednisolone was of no benefit so the drug was tapered. Hydroxyurea was started and continued for

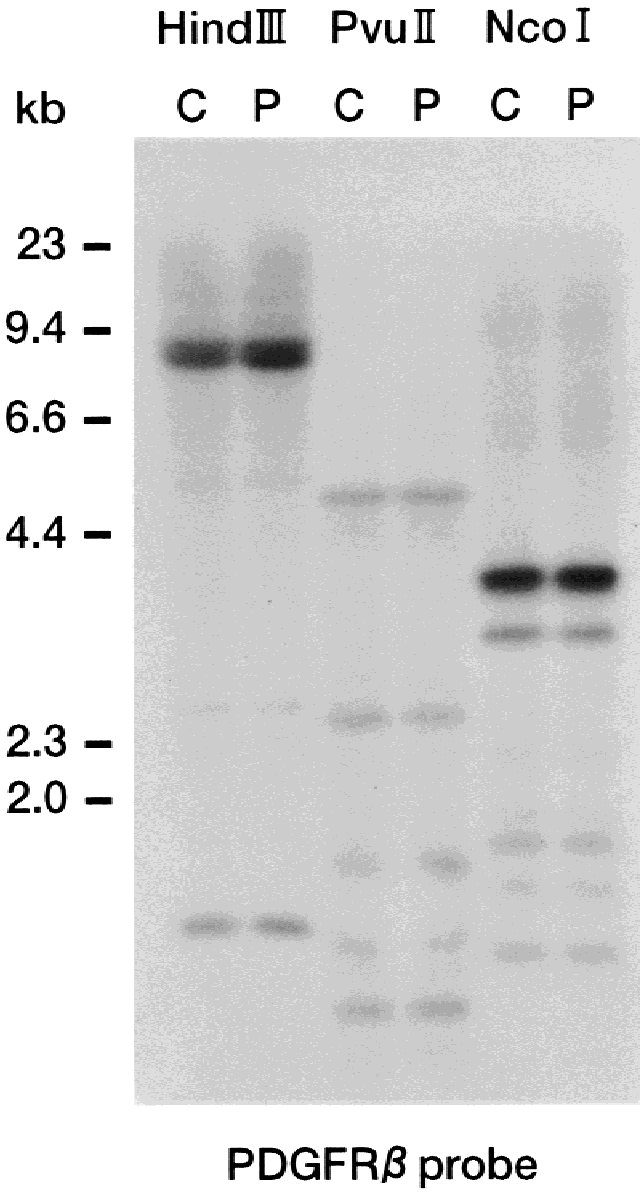

Fig. 2. Southern blot analysis of DNA from the patient before treatment with interferon-alpha. Genomic DNA from the patient (lane P) and control human placental DNA (lane C) were digested with the restriction enzymes *Nco* I, *Pvu* II, and *Hind* III and hybridized with the PDGFRβ cDNA probe. Size markers are *Hind* III-digested lamda phage DNA (kb).

about 1 year, but anemia developed and blood transfusions were required. Therapy was changed to etoposide according to published data [3]. Erythropoietin was started (Epogen, Chugai, Tokyo, Japan) at 6,000 to 9,000 U three times weekly to support erythropoiesis. In August 1991, the patient suffered a left occipital cerebrovascular accident but recovered with a minimal residual visual deficit, and warfarin therapy was started. In August 1992, the patient developed a generalized herpes infection, and etoposide was discontinued in December 1992. After informed consent was obtained, natural

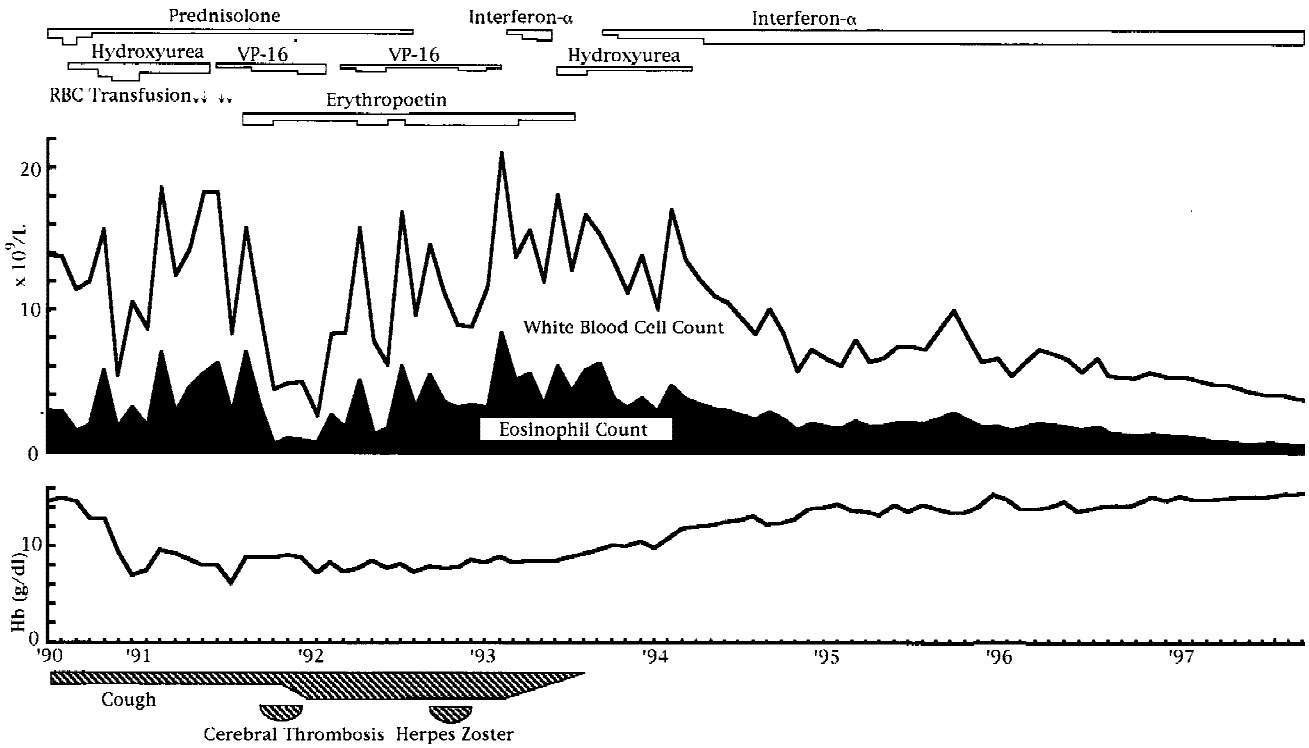

Fig. 3. White blood cell count and total eosinophil count during treatment with interferon-alpha.

IFN-α (Sumiferon, Sumitomo, Osaka, Japan) was started at a dose of 1.5×10^6 U per day subcutaneously in late 1992. This dosage was increased by 1.5×10^6 U per day every 4 weeks as tolerated. IFN-α was temporarily replaced by hydroxyurea and resumed in June 1993. The dose of IFN-α was increased to 6×10^6 U over the next 6 months. In the first 12 months of reinstitution of interferon therapy, the white blood cell count decreased to 6×10^9/1, the total eosinophil count averaged about 1.5×10^9/1, and anemia resolved. The bone marrow became normocellular with 16% mature eosinophils and a normal karyotype. Chromosomal analysis of the bone marrow from the time of diagnosis up to the time of cytogenetic remission was serially studied by Q-banding technique and the behavior of this populations is best seen in Table I. The patient had a dramatic clinical response following administration of IFN-α with marked improvement in congestive heart failure. The cardiothoracic ratio decreased from 58 to 50%. The patient has since been clinically stable and fully functional and remains well on IFN-α 6×10^6 U daily at this writing.

TABLE I. Cytogenetic Pattern Before and After IFN-α Treatment*

Date	Source	No. of mitoses examined	t(3;9;5)	Diploid (46,XY)	IFN-α
90.7.05	BM	30	30	0	−
90.7.25	PB(+PHA)	30	0	30	−
91.5.24	BM	29	25	4	−
93.6.15	BM	30	21	9	+
94.1.20	BM	38	13	25	+
94.10.6	BM	30	0	30	+
96.12.12	BM	30	0	30	+

*BM, bone marrow; PB, peripheral blood; PHA, phytohemaggrutinin; IFN-α, interferon-alpha.

DISCUSSION

The similarity between HES and other myeloproliferative syndromes such as CML has been noted in the past [4]. HES is difficult to distinguish from other hypereosinophilic states, and the diagnosis is often made by exclusion. When evidence supporting a myeloproliferative disorder is found, the diagnosis of HES is no longer appropriate, and patients with a clonal cytogenetic abnormality should be classified as having eosinophilic leukemia [1].

It is not likely that the case presented here meets the diagnostic criteria of chronic myelogenous leukemia, myelodysplastic syndrome, or lymphoid malignancies. Given the monotypic result on cytogenetic analysis, the mitotic feature of eosinophils in the patient's bone marrow strongly suggests that precursors of an eosinophilic series have the unique translocation, t(3;9;5)(q25;q34;q33). The long clinical course and bone marrow with a predominance of eosinophilic precursors but without an excess of blast cells and with other normal

hemopoietic cells, gave a presumptive diagnosis of chronic eosinophilic leukemia even though conclusive proof of the neoplastic nature of the eosinophils may be lacking. Firmer evidence will be provided when fluorescence in situ hybridization demonstrates a cytogenetic abnormality in eosinophils [5], or when clonal cytogenetic abnormalities are demonstrated in eosinophil colonies [6].

In view of the beneficial effects of IFN-α on CML [2], recent studies have advocated the use of IFN-α in some patients with HES and eosinophilic leukemias, and the effectiveness of IFN-α was summarized in a series of recent publications [7]. IFN-α is known to suppress eosinophilic colony growth in vitro [8], suggesting the existence of a regulatory system of growth factors affecting eosinophil proliferation [9]. The dramatic improvement in cardiopulmonary function while the absolute eosinophil count still remained about $1.5 \times 10^9/1$ suggests that the beneficial action of IFN-α in our patient may be mediated through effects on the mature eosinophil. Direct effects of IFN-α on eosinophils have been observed, including inhibition of chemotaxis and decreased production of hydrogen peroxide, neurotoxin, and peroxidase in response to stimulation [10,11].

Various chromosomal abnormalities have been described in blood disorders with eosinophilia [7,12,13]. They are classified into three types: trisomy 8, and abnormalities in 12p13 and 5q31–33. An extra chromosome 8 has been described in association with eosinophilic leukemia or HES but is also a common finding in myeloid disorders. In 44 cases of chronic eosinophilic leukemia analysed with chromosomal banding, 33 patients had unique abnormalities: a constant region affected in these translocations was a breakpoint in the long arm of chromosome 5 and/or in the short arm of chromosome 12 [1]. Among previously reported patients with HES or eosinophilic leukemia treated with IFN-α, cytogenetic remission was obtained in one case: t(5;9)(q32;q33) [7]. Our case is the first to be reported of chronic eosinophilic leukemia with a previously undescribed chromosomal abnormality 46,XY,t(3;9;5)(q25;q34;q33) that showed clinical and cytogenetic remission after treatment with INF-α.

The mechanism responsible for proliferation of eosinophils in cases of eosinophilic leukemia with demonstrable cytogenetic lesions is unknown. Three cytokines that are known to affect proliferation and differentiation of eosinophil progenitors include IL-3, IL-5, and GM-CSF [14]. Plasma levels of GM-CSF and IL-3 in our patient were normal, but the blood sample that was available to measure the IL-5 level 1 year after reinstitution of interferon therapy showed an increase of this cytokine. The genes encoding for these cytokines reside on the long arm of chromosome 5 (5q23–31) [15–17], and the gene encoding the tyrosine kinase domain of the PDGFRβ is located on 5q33 [18]. The translocation in which the *ets*-like gene, *tel*, was fused to PDGFRβ was first identified in chronic myelomonocytic leukemia with t(5;12) chromosomal translocation [11]. The *tel*-PDGFRβ fusion causes constitutive tyrosine kinase activation of PDGFRβ, resulting in the promotion of cellular proliferation and activation of the signal transduction pathway [19,20]. In our case, however, we could not detect this rearrangement when the PDGFRβ gene was probed with the full length of human PDGFRβ cDNA [21]. Some other unknown mechanism should be considered in such variant cases of t(5;12) translocation.

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